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2018-03

Aly , A A , El-Sheref , E M , Mourad , A-F E , Brown , A B , Bräse , S , Bakheet , M E M & Nieger , M 2018 , ' Synthesis of spiro[indoline-3,4'-pyrano[3,2-c]quinolone]-3'-carbonitriles ' , Monatshefte für Chemie , vol. 149 , no. 3 , pp. 635-644 . https://doi.org/10.1007/s00706-017-2078-6

http://hdl.handle.net/10138/309240
https://doi.org/10.1007/s00706-017-2078-6

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acceptedVersion

Synthesis of spiro(indoline-3,4'-pyrano[3,2-*c*]quinoline)-3'-carbonitriles

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Received: ……/Accepted …

Abstract. Quinoline-2,4-diones reacted with 2-(2-oxo-1,2-dihydroindol-3-ylidene)malononitrile in pyridine to give 2'-amino-2,5'-dioxo-5',6'-dihydro-spiro(indoline-3,4'-pyrano[3,2-*c*]quinoline)-3'-carbonitriles in good to excellent yields. The structures of all new products were proven using one- and two-dimensional NMR, IR, and mass spectral data, and in four cases X-ray structural analyses. The possible mechanism for the reaction is also discussed.

Keywords. Quinolin-2,4-diones ● Isatinemalononitrile ● New spiro-compounds ● X-ray analysis

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Introduction

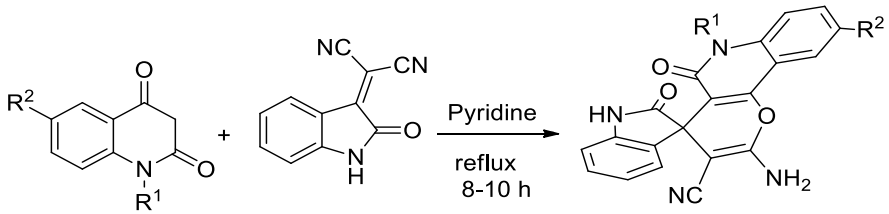
Great efforts have been made to synthesis, characterize and investigate the biological activities of these compounds. They possess very promising biological activities as anticonvulsant [1-3], antibacterial [4], anti-Alzheimer [5], antimicrobial [6], anti-dermatities [7], anticancer [8], and pain-relief [9], in addition to their medical, agricultural and industrial uses [10-12]. In general, spiro-compounds have also drawn tremendous interest of researchers in synthetic organic chemistry and medicinal chemistry due to their extensive applications in biology and pharmacology [13]. Indeed, quinolones represent a class of fused ring systems with interesting essential role in the construction of many bioactive compounds, that they can exhibit antibacterial [14], antifungal [14], anti-tumor [15], anticancer [16], antituberculosis [16], antibiotic [17], anti-microbial [18], anti-inflammatory activities [18]. However, they have herbicides [19], and insecticides agent [20]. On the other hand, isatinemalononitrile is a scaffold for the synthesis of fused spiro-compounds [21], many of its derivatives are potent inhibiting caspase [22]. The heterocyclic spiro-oxindole framework is an important structural motif in natural products which can act as potent non-peptide inhibitors [23]. These compounds are very interesting: they can act as antibacterial and anti-HIV agents [24], and exhibit biological activities [25], such as antitumor and anticancer [26] and cellularevaluation [27].

To extend the knowledge around the new spiro-compounds, we focused our searches to synthesis a new class of spiro-compounds which we expect they will have importance activities in medicinal and industrial area. Previously, Aly *et al.*, synthesized fused spiro-pyranoindanoparacyclophanes [28], spiro-pyridazinocyclohexadiene as well as spiro-thiadiazolopyrimidinocyclo-hexadiene derivatives [29], and spiro(indole-3,3′-[1,2,4]-triazol)-2(1*H*)-ones [30].

For all these reasons, synthesis of spiro-indolinepyrano[3,2-*c*]quinolines continues to attract interest. Herein we describe the synthesis of a class of new spiro-compounds, *via* the reaction of quinoline-2,4-(1*H*,3*H*)-diones **1a-f** with 2-(2-oxo-1,2-dihydroindol-3-ylidene)malononitrile (**2**).

2. Results and Discussion

Refluxing equimolar amounts of 1,6-disubstituted quinoline-2,4-(1*H*,3*H*)-diones **1a-f** with 2-(2-oxo-1,2-dihydroindol-3-ylidene)-malononitrile (**2**) in dry pyridine solution led to the formation of spiro(indoline-3,4'-pyrano[3,2-*c*]quinoline)-3'-carbonitriles **3a-f** in 85-92% yields (Scheme 1).



1a-f **2** **3a-f** (85-92%)

3a (85%), **3b** (92%), **3c** (90%), **3d** (88%), **3e** (87%), **3f** (86%)

1,3: a, $R^1 = R^2 = H$; **b**, $R^1 = H$, $R^2 = Cl$; **c**, $R^1 = H$, $R^2 = Br$; **d**, $R^1 = H$, $R^2 = Me$; **e**, $R^1 = Me$, $R^2 = H$; **f**, $R^1 = Et$, $R^2 = H$

Scheme 1. Synthesis of spiro-compounds **3a-f** *via* reaction between quinoline-2,4-diones **1a-f** and **2**.

To confirm the structures of all the obtained products, elemental analyses, IR, NMR (^1H, ^{13}C, 2D NMR, ^{15}N) and mass spectra were performed; these and elemental analyses were in good agreement with the assigned structures. To illustrate the structure elucidation, we choose a representative example, 2'-amino-6'-methyl-2,5'-dioxo-5',6'-dihydrospiro(indoline-3,4'-pyrano[3,2-*c*]quinoline)-3'-carbonitrile (**3e**) (Table 1). According to elemental analysis and mass spectrometry, compound **3e** has the gross formula $C_{21}H_{14}N_4O_3$, resulting from combination of one molecule of *N*-methylquinoline-2,4-dione (**1e**) with one molecule of **2**. Our first impression was that the reaction might give either **3e**, **4**, or **5** (Figure 1). The NMR data appear in Table 1.

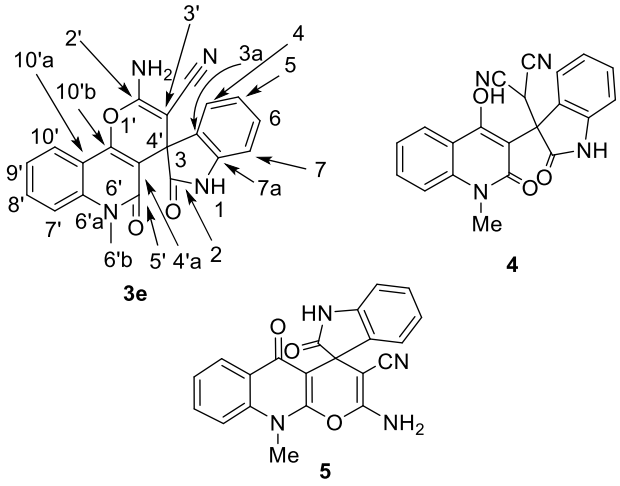


Figure 1. Alternative expected structure of compound **3e**.

Table 1. NMR assignment of compound **3e** ^{1}H NMR

^{1}H NMR	^{1}H-^{1}H COSY	Assig.
10.54 (bs; 1H)		NH-1
8.07 (d, *J* = 8.0; 1H)	7.76	H-10'
7.76 ("dt", J_t = 7.3, J_d = 1.1; 1H)	8.07,7.59,7.45	H-8'
7.59 (d, *J* = 8.6; 1H)	7.76,7.45	H-7'
7.48 (bs; 2H)		NH_2
7.45 (t, *J* = 7.7; 1H)	7.76	H-9'
7.18 (t, *J* = 7.6; 1H)	7.02,6.88,6.85	H-6
7.02 (d, *J* = 7.2; 1H)	6.88,6.85	H-4
6.88 (t, *J* = 7.5; 1H)	7.18,7.02	H-5
6.85 (d, *J* = 7.8; 1H)	7.18,7.02	H-7
3.49 (s; 3H)		CH_3

^{15}N NMR	HSQC	HMBC	Assig.
140.5		7.59,3.49	N-6'
136.3	10.54	6.85	N-1
75.2	7.48		NH_2

^{13}C NMR	HSQC	HMBC	Assig.
177.81		10.54	C-2
158.87,158.72		7.48,3.49	C-5',2'
151.46		8.07,7.59	C-10'a
142.48		10.54,7.18,7.02,6.85	C-7a
138.64		8.07,7.76,3.49	C-6'a
134.25		10.54,6.88,6.85	C-3a
132.23	7.76	8.07,7.59	C-8'
128.28	7.18	7.02,6.88	C-6
123.40	7.02	7.18	C-4
122.41,122.36	8.07,7.45	7.76,7.59	C-9',10'
121.66	6.88	6.85	C-5
117.43			CN
115.03	7.59	3.49	C-7'
112.27		7.59,7.48	C-10'b
109.18	6.85	6.88	C-7
106.48			C-4'a
57.22		7.48	C-3'
48.15		10.54,7.02,6.88,6.85	C-3,4'
29.19	3.94		CH_3

The ^{1}H spectrum consists of two four-spin coupled networks in the aromatic region, two broadened singlets (1H and 2H), and a methyl singlet. The ^{13}C spectrum has 21 lines, consistent with **3e** but excluding structure **4**; eighteen are in the normal sp^2region between δ_C = 100-160 ppm. Three nitrogen atoms were observed, all but the nitrile. One of the aromatic proton systems appears between δ_H = 8.07-7.45 ppm and the other between δ_H = 7.18-6.85 ppm. To differentiate the two aromatic systems, the nearby nitrogen atoms are convenient starting points. The sp^2 nitrogen at δ_N = 136.3 ppm, gives HSQC correlation with the 1H broadened singlet at δ_H = 10.54 ppm; these two signals are assigned as N-1 and the attached proton. Two carbons at δ_C = 142.48 and 134.25 ppm give HMBC correlation both with H-1 and with protons in the system δ_H = 7.18-6.85 ppm, and are assigned as C-7a and C-3a in some order. H-1 also gives HMBC correlation with the carbonyl carbon at δ_C = 177.81 ppm, which is assigned as C-2, and the sp^3 carbon at δ_C = 48.15 ppm, assigned as the spiro carbon C-3,4'. The spiro carbon gives HMBC correlation with the proton doublet at δ_H = 7.02 ppm, assigned as H-4 (three bonds away from the spiro carbon); the attached carbon appears at δ_C = 123.40 ppm. H-4 also gives HMBC correlation with the aforementioned carbon at δ_C = 142.48 ppm, which is assigned as C-7a (three bonds away from H-4); by subtraction, the carbon at δ_C = 134.25 ppm, is assigned as C-3a. Also, C-7a gives HMBC correlation with a proton triplet at δ_H = 7.18

ppm, assigned as H-6; this is another three-bond correlation, and the attached carbon appears at δ_C = 128.28 ppm. The remaining triplet and doublet in this aromatic proton system appear at δ_H = 6.88 and 6.85 ppm, and are assigned as H-5 and H-7 respectively; the attached carbons appear at δ_C = 121.66 and 109.18. The other sp^2 nitrogen, at δ_N = 140.5, gives HMBC correlation with an aromatic doublet at δ_H = 7.59 ppm, and a methyl singlet at δ_H = 3.49 ppm. This nitrogen is assigned as N-6' and the methyl singlet as H-6'b; the carbon attached to H-6'b appears at δ_C = 29.19 ppm, and is assigned as C-6'b. The proton at δ_H = 7.59 is assigned as H-7', which is three bonds from N-1; its attached carbon appears at δ_C = 115.03 ppm, and gives HMBC correlation with H-6'b. On the other hand, H-6'b also gives HMBC correlation with a carbon at δ_C = 138.64, assigned as C-6'a; this is a three-bond correlation. C-6'a also gives HMBC correlation with an aromatic doublet at δ_H = 8.07 and a triplet at δ_H = 7.76 ppm, assigned as H-10' and H-8' respectively; the attached carbons appear at δ_C = 122.41 or 122.36, and at δ_C = 132.23 ppm. The remaining triplet in this ring appears at δ_H = 7.45 ppm, and is assigned as H-9'; its attached carbon is the other of δ_C = 122.41 and 122.36. A carbon at δ_C = 151.46 ppm gives HMBC correlation with H-10' and H-8', and is assigned as C-10'a. The remaining carbons at δ_C = 158.87, 158.72, 117.43, 112.27, 106.48, and 57.22 ppm, must be C-2', 3', CN, 4'a, 5', and 10'b. The two carbons at δ_C = 158.87 and 158.72 ppm, give HMBC correlation with H-6'b and the

broadened 2H signal at δ_H = 7.48 ppm; these carbons are assigned as C-2' (which should correlate with the NH_2 group) and C-2' (which is three bonds from H-6'b).The broadened protons just mentioned give HSQC correlation with the *sp3* nitrogen at δ_N = 75.2, assigned as NH_2. The upfield carbon at δ_N = 57.22 gives HMBC correlation with N**H**$_2$, and is assigned as C-3'; its upfield shift is attributed to the observed trends in δ values for C-atoms in push-pull alkenes [31,32]. The carbon at δ_C = 112.27 gives HMBC correlation with H-7' and N**H**$_2$; it is assigned as C-10'b, which is four bonds from each of these protons. The remaining carbons appear at δ_C = 117.43 and 106.48 ppm, and must be CN and C-4'a; based on chemical shifts, the downfield of the two is assigned as the nitrile carbon-CN and the upfield carbon as C-4'a.

As another example, we choose **3a**. In its IR spectrum, bands at 3207-3372 cm^{-1} can be assigned to NH_2 and NH groups, and strong absorption at 2205 cm^{-1} is characteristic of a cyano group. Several peaks appear at 1725-1642 and 1600, 1596 cm^{-1} as a result of C=O stretching and skeletal vibration of the aryl group, respectively. The structure of compound **3a** was previously prepared [21f], however its detailed NMR data were not extensively reported. The NMR spectra of **3a** (Table 2) are very similar to those of **3e**, except that instead of the N-methyl group, there is a broadened 1H singlet at δ_H = 11.75 ppm, assigned as NH-6. This proton gives HSQC correlation with N-6, and gives HMBC correlation with C-2',

C-3'. C-3,4', C-5', C-9', C-10', and C-10'b. All these correlations are over four bonds or fewer; the structure assignment of **3a** is unambiguously confirmed by an X-ray crystal structure (Figure 2).

Table 2. NMR assignments of compound **3a** ^{1}H NMR

^{1}H NMR	^{1}H-^{1}H COSY	Assig.
11.75 (bs; 1H)		NH-6'
10.53 (bs; 1H)		NH-1
7.96 (d, *J* = 7.8; 1H)	7.63	H-10'
7.63 (ddd, *J* = 8.3, 7.2, 1.1; 1H)	7.96, 7.36	H-8'
7.46 (bs; 2H)		NH_2
7.36 (d, *J* = 8.1; 1H)	7.63	H-7'
7.34 (dd, *J* = 7.5, 7.5; 1H)	7.63	H-9'
7.19 (ddd, *J* = 7.6, 7.6, 0.7; 1H)	7.04, 6.90, 6.84	H-6
7.04 (d, *J* = 7.3; 1H)	7.19, 6.90, 6.84	H-4
6.90 (dd, *J* = 7.5, 7.4; 1H)	7.19, 7.04, 6.84	H-5
6.84 (d, *J* = 7.7; 1H)	7.19, 7.04, 6.90	H-7

^{15}N NMR	HSQC	HMBC	Assig.
146.2	11.75	7.36	N-6'
136.3	10.54	6.84	N-1
75.3	7.46		NH_2

^{13}C NMR	HSQC	HMBC	Assig.
177.83		10.54	C-2
159.40,158.94		11.75,7.48	C-5',2'
152.39		11.75,7.96,7.48,7.36	C-10'a
142.37		10.54,7.19,7.04,6.90,6.84	C-7a
137.82		11.75,7.96,7.63,7.34	C-6'a
134.31		10.54,7.19,6.90,6.84	C-3a
131.77	7.63	7.96	C-8'
128.28	7.19	7.04,6.90	C-6
123.46	7.04	7.19	C-4
122.16	7.34	7.63,7.36	C-9'
121.91	6.90	6.84	C-5
121.68	7.96	7.63	C-10'
117.48			CN
115.37	7.36	11.75,7.34	C-7'
111.55		11.75,7.63,7.36,7.34	C-10'b
109.22	6.84	7.18,7.04,6.90	C-7
106.99		11.75	C-4'a
57.18		7.48	C-3'
47.75		11.75,10.54,7.04,6.84	C-3,4 '

The spectra of the *N*-ethyl compound **3f** are essentially identical to those of **3e**; because spiro carbon C-14 is a stereogenic center, protons H-101 are diastereotopic [33], and the *N*-ethyl group appears as an ABX_3 system (**3f**). The structure

assignment of **3f** is unambiguously confirmed by an X-ray crystal structure; the crystal contains one equivalent of DMF, the recrystallization solvent (Fig. 5).

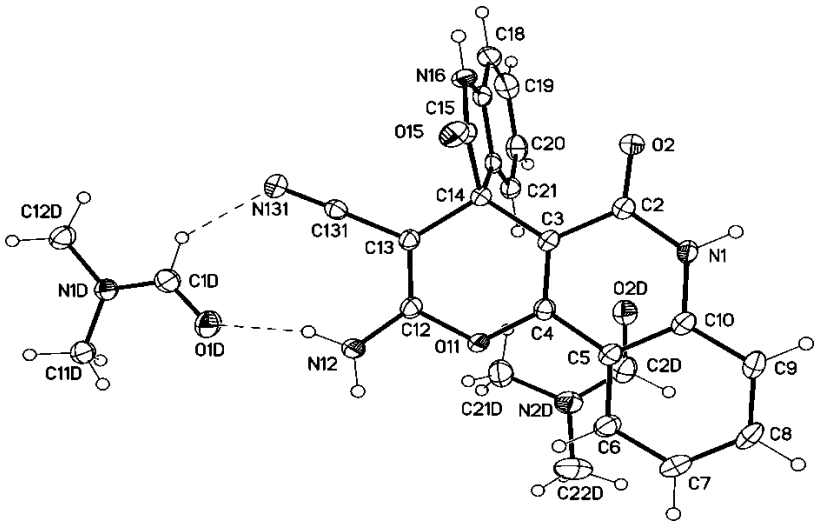


Figure 2. Molecular structure of **3a**.**DMF** 2'-Amino-2,5'-dioxo-5',6'-dihydrospiro(indoline-3,4'-pyrano-[3,2-c]quinoline)-3'-carbonitrile.DMF (1:1). Displacement parameters are drawn at 50% probability level.

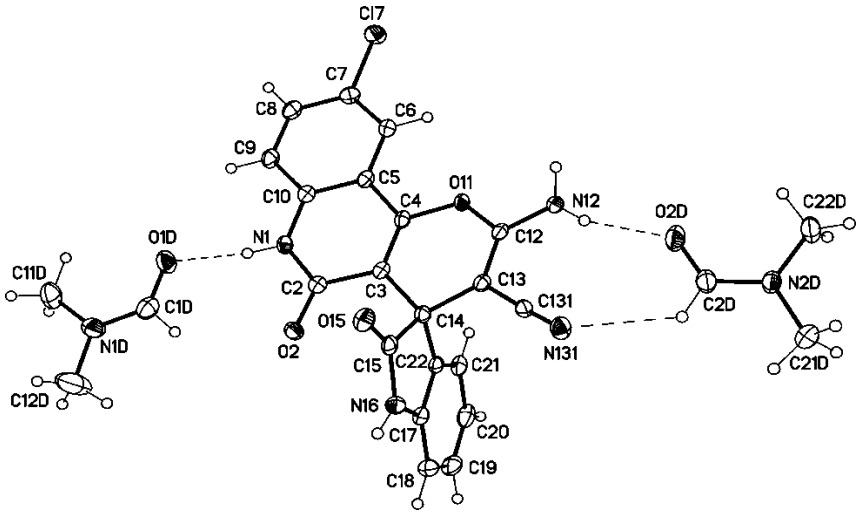


Figure 3. Molecular structure of **3b**.**2DMF**. (2'-amino-9'-chloro-2,5'-dioxo-5',6'-dihydrospiro[indoline-3,4'-pyrano[3,2-*c*]quino-line)-3'-carbonitrile.2DMF. Displacement parameters are drawn at 50% probability level.

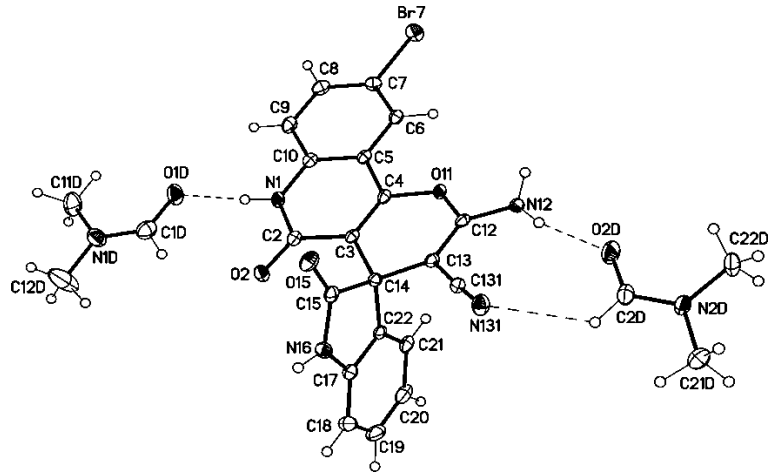


Figure 4. Molecular structure of **3c.2DMF**. 2'-amino-9'-bromo-2,5'-dioxo-5',6'-dihydrospiro(indoline-3,4'-pyrano[3,2-c]quino-line)-3'-carbonitrile.2DMF (1:2). Displacement parameters are drawn at 50% probability level.

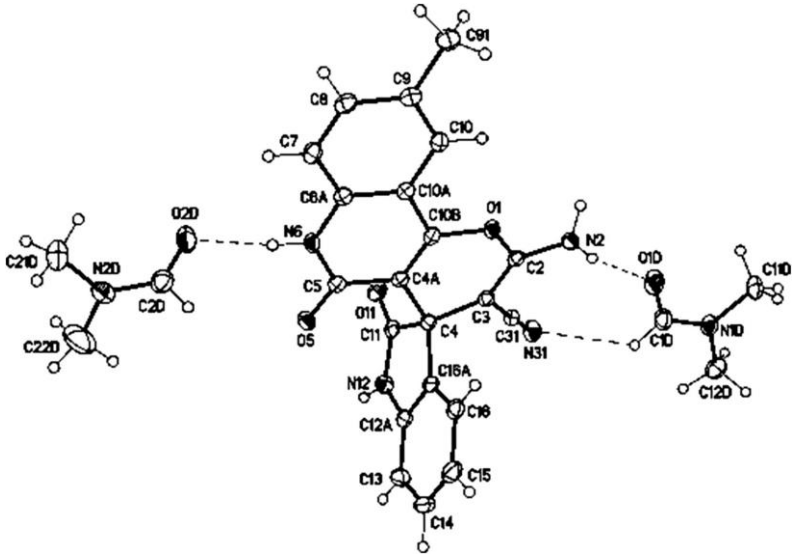


Figure 4. Molecular structure of **3d DMF**. Displacement parameters are drawn at 50% probability level

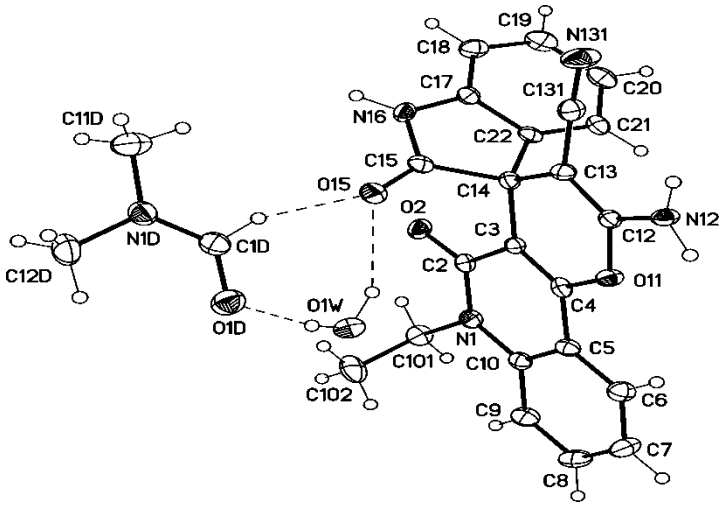
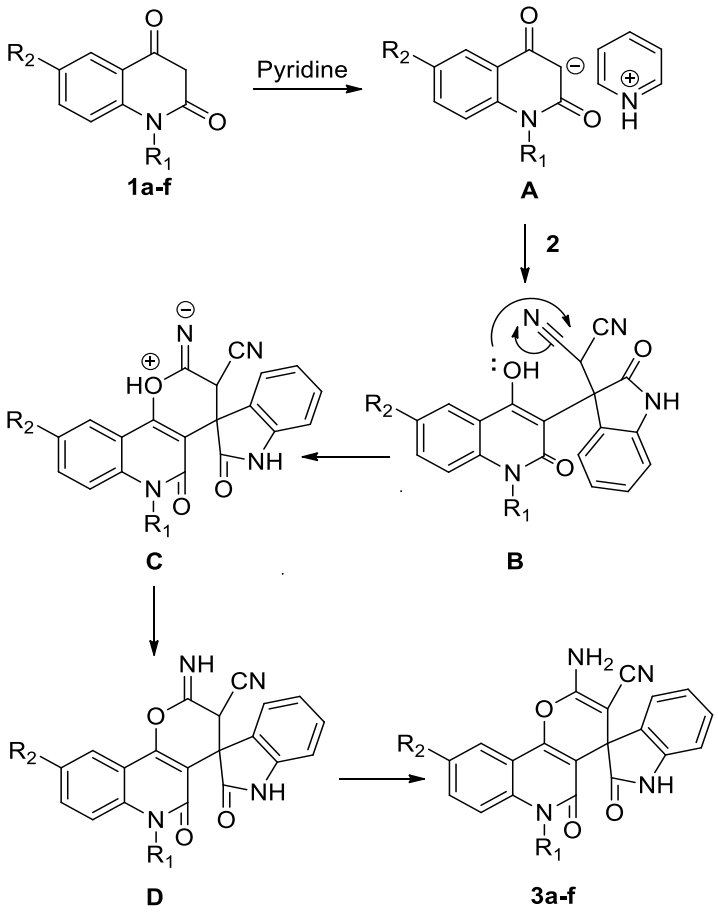


Figure 5. Molecular structure of **3f.DMF** 2'-Amino-6'-ethyl-2,5'-dioxo-5',6'-dihydrospiro(indoline-3,4'-pyrano[3,2-*c*]quinoline)-3'-carbonitrile.DMF (1:1). Displacement parameters are drawn at 50% probability level.

From the 2D NMR correlations the structures **5** can be excluded, but another confirmation was made by using X-ray analysis as shown in Figures 2-5.

We propose the mechanism shown in Scheme 2. Conjugate addition of **1** to **2**, catalyzed by base, would give intermediate **B**. Cyclization of **B** would then occur to give intermediate **C** and, after proton transfer, finally give products **3a-f** (Scheme 2).



Scheme 2. Suggested mechanism for formation of products **3a-f**

Most indicative is the ring conformation in crystals of compounds **3a-c** and **3f** are distorted under the effect of intermolecular interactions, as is evidenced by short intermolecular contacts. The complexes are stabilized by intermolecular N-H···O and C-H···O hydrogen bonds between the hydrogen atoms situated inside the cavity of the macrocycle, the oxygen and nitrogen atoms of the dimethylformamide and water molecules.

3. Conclusion

Reaction of 2,4-quinolinediones with isatinemalononitrile yields spiro compounds in good yields; structures were established by solution-phase spectroscopy and X-ray crystallographic analysis.

4. Experimental

4.1. General

NMR spectra were measured in DMSO-d_6 on a Bruker AV-400 spectrometer (Bruker BioSpin Corp., Billerica, MA, USA) (400.13 MHz for ^{1}H, 100.13 MHz for ^{13}C, and 40.55 MHz for ^{15}N) at Florida Institute of Technology, USA. The ^{1}H and ^{13}C chemical shifts are given relative to internal standard TMS; ^{15}N shifts are reported versus external liquid ammonia. For preparative thin layer chromatography (PLC), glass plates (20 x 48 cm) were covered with a slurry of silica gel Merck PF254 and air dried and developed using the solvents listed. Zones were detected by quenching of indicator fluorescence upon exposure to 254 nm UV light.

Elemental analyses were carried in the National Research Center, Dokki, Cairo, Egypt. Mass spectrometry was performed by electron impact at 70 eV, with a Finnigan Mat 8430 spectrometer in the National Research center, Dokki, Cairo, Egypt. IR spectra using KBr pellets, were run on a FT-IR (Bruker), Minia University, El-Minia, Egypt.

4.2. Starting materials

Quinoline-2,4-diones **1a-f** were prepared according to the literature [34].

4.3. X-ray crystal structure determination

The single-crystal X-ray diffraction study was carried out on a Bruker D8 Venture diffractometer with Photon100 detector at 123(2) K using Cu-Kα radiation (**3a**, **3b**, **3f**, λ = 1.54178 Å) or Mo-Kα radiation (**3c**, λ = 0.71073 Å). Direct Methods (SHELXS-97) [35] were used for structure solution and refinement was carried out using SHELXL-2014 (full-matrix least-squares on F^2) [36]. Hydrogen atoms were localized by difference electron density determination and refined using a riding model (H(N) and H(O) free. Semi-empirical absorption corrections were applied. For **3a** and **3c** an extinction correction was applied.

3a: colourless crystals, $C_{20}H_{12}N_4O_3 \cdot 2(C_3H_7NO)$, M_r = 502.53, crystal size 0.16 × 0.10 × 0.08 mm, monoclinic, space group $P2_1/c$ (No. 14), a = 10.8078(3) Å, b = 21.4185(6) Å, c = 11.0025(3) Å, β = 106.635(1)°, V = 2440.34(12) Å^3, Z = 4, ρ = 1.368 Mg/m^{-3}, μ(Cu-K$_\alpha$) = 0.805 mm^{-1}, F(000) = 1056, $2\theta_{max}$ = 144.4°, 26760

reflections, of which 4805 were independent (R_{int} = 0.042), 351 parameters, 4 restraints, R_1 = 0.038 (for 4182 I > 2σ(I)), wR_2 = 0.097 (all data), S = 1.06, largest diff. peak / hole = 0.297 / -0.206 e Å^{-3}.

3b: colourless crystals, $C_{20}H_{11}ClN_4O_3 \cdot 2(C_3H_7NO)$, M_r = 536.97, crystal size 0.20 × 0.16 × 0.10 mm, monoclinic, space group $P2_1/c$ (No. 14), a = 11.0744(4) Å, b = 21.3368(8) Å, c = 11.1215(4) Å, β = 107.884(1)°, V = 2500.94(16) Å^3, Z = 4, ρ = 1.426 Mg/m^{-3}, μ(Cu-K$_\alpha$) = 1.784 mm^{-1}, F(000) = 1120, $2\theta_{max}$ = 144.2°, 21016 reflections, of which 4915 were independent (R_{int} = 0.027), 359 parameters, 4 restraints, R_1 = 0.036 (for 4445 I > 2σ(I)), wR_2 = 0.097 (all data), S = 1.03, largest diff. peak / hole = 0.320 / -0.290 e Å^{-3}.

3c: colourless crystals, $C_{20}H_{11}BrN_4O_3 \cdot 2(C_3H_7NO)$, M_r = 581.43, crystal size 0.25 × 0.20 × 0.15 mm, monoclinic, space group $P2_1/c$ (No. 14), a = 11.0650(5) Å, b = 21.4404(11) Å, c = 11.2361(5) Å, β = 108.179(2)°, V = 2532.6(2) Å^3, Z = 4, ρ = 1.525 Mg/m^{-3}, μ(Mo-K$_\alpha$) = 1.673 mm^{-1}, F(000) = 1192, $2\theta_{max}$ = 55.2°, 39390 reflections, of which 5834 were independent (R_{int} = 0.024), 360 parameters, 4 restraints, R_1 = 0.026 (for 5290 I > 2σ(I)), wR_2 = 0.064 (all data), S = 1.05, largest diff. peak / hole = 0.483 / -0.572 e Å^{-3}.

3f: colourless crystals, $C_{22}H_{16}N_4O_3 \cdot C_3H_7NO \cdot H_2O$, M_r = 475.50, crystal size 0.16 × 0.10 × 0.04 mm, triclinic, space group P-1 (No. 2), a = 9.6532(5) Å, b = 11.3165(6) Å, c = 12.6497(6) Å, α = 110.480(3)°, β = 102.514(3)°, γ = 107.481(3)°,

V = 1151.89(11) Å^3, Z = 2, ρ = 1.371 Mg/m^{-3}, μ(Cu-K$_\alpha$) = 0.807 mm^{-1}, F(000) = 500, $2\theta_{max}$ = 136.4°, 11728 reflections, of which 4141 were independent (R_{int} = 0.037), 333 parameters, 5 restraints, R_1 = 0.055 (for 3170 I > 2σ(I)), wR_2 = 0.155 (all data), S = 1.05, largest diff. peak / hole = 0.354 / -0.254 e Å^{-3}.

CCDC-1519994 (**3a**), CCDC-1519995 (**3b**), CCDC-1519996 (**3c sb897_hy ME-86**) and CCDC-1519997 (**3f**) contain the supplementary crystallographic data for this paper. These data can be obtained free of charge from The Cambridge Crystallographic Data Centre *via* www.ccdc.cam.ac.uk/data_request/cif.

4.4. General Procedure

*4.5. Reaction of quinoline-2,4-diones **1a-f** with **2***

A 100 mL round-bottom flask was flame-dried, a mixture of **1a-f** (1 mmol), **2** (1 mmol) and 20 ml dry pyridine was refluxed for 8-10 h with stirring (the reaction was followed by TLC analysis). After the reaction's completion, solvent was then removed under vacuum and the residue was separated. The solid residue undergoes recrystallization from dry DMF to give pure crystals of spiro-compounds **3a-f**.

2'-Amino-2,5'-dioxo-5',6'-dihydrospiro(indoline-3,4'-pyrano-[3,2-*c*]quinoline)-3'-carbonitrile (3a). Colorless crystals (303 mg, 85 %), M.p.: 298-300°C; IR (KBr, cm^{-1}): $\bar{\nu}$ = 3372-3207 (NH, NH$_2$), 3099 (Ar-H), 2205 (CN), 1725, 1672, 1642 (C=O), 1600, 1596 (Ar-C=N, Ar-C=C); NMR (DMSO-d_6) (Table 1); MS (Fab, 70 eV, %): m/z = 356

(M$^+$, 100). *Anal. Calcd. For* $C_{20}H_{12}N_4O_3$: C, 67.41; H, 3.39; N, 15.72; Found: C, 67.55; H, 3.22; N, 15.66.

2'-Amino-9'-chloro-2,5'-dioxo-5',6'-dihydro-spiro(indoline-3,4'-pyrano[3,2-*c*]quinoline)-3'-carbonitrile(3b). Colorless crystals (360 mg, 92%), M.p.: 320-2^{o}C; IR (KBr, cm^{-1}): $\overline{\nu}$ = 3350-3196 (NH, NH_2), 3031 (Ar-CH), 2928 (Ali-CH), 2193 (CN), 1716, 1669, 1654 (C=O), 1605, 1593 (Ar-C=N, Ar-C=C); ^{1}H NMR (400 MHz, DMSO-d_6): δ_H = 11.88 (bs, 1H; NH-6'), 10.55 (bs; 1H; NH-1), 8.00 (d, *J* = 2.3; 1H, H-10'), 7.68 (dd, *J* = 8.8, 2.4; 1H, H-8'), 7.47 (bs; 2H, NH_2),7.37 (d, *J* = 8.8; 1H, H-7'), 7.19 (ddd, *J* = 7.6, 7.6, 0.7; 1H, H-6), 7.05 (d, *J* = 7.3; 1H, H-4), 6.90 (dd, *J* = 7.4, 7,4; 1H, H-5), 6.83 ppm (d, *J* = 7.7; 1H, H-7) ppm; ^{13}C NMR (100 MHz, DMSO-d_6): δ_C = 177.58 (C-2), 159.18, 158.77 (C-5',2'), 151.44 (C-10'a), 142.33 (C-7a), 136.55 (C-6'a), 134.06 (C-3a), 131.71 (C-8'), 128.38 (C-6), 126.31 (C-9'), 123.56 (C-4), 121.71 (C-5), 121.23 (C-10'), 117.48, 117.30 (CN,C-7'), 111.55 (C-10'b), 109.24 (C-7), 107.98 (C-4'a), 57.09 (C-3'), 47.74 ppm (C-3,4') ppm; ^{15}N NMR (DMSO-d_6): δ_N = 146.0 (N-6'), 136.2 (N-1), 74.8 (NH_2) ppm; MS (Fab, %): *m/z* = 391 (M+1, 28), 390 (M$^+$, 58). *Anal. Calcd. for* $C_{20}H_{11}ClN_4O_3$: C, 61.47; H, 2.84; Cl, 9.07; N, 14.34; Found: C, 61.33; H, 3.01; Cl, 9.22; N, 14.29.

2'-Amino-9'-bromo-2,5'-dioxo-5',6'-dihydro-spiro(indoline-3,4'-pyrano[3,2-*c*]quinoline)-3'-carbonitrile (3c). Colorless crystals (390 mg, 90%), M.p.: > 360 ^{o}C; IR (KBr, cm^{-1}): $\bar{\nu}$ = 3365-3195 (NH, NH_2), 3044 (Ar-CH), 2930 (Ali-CH), 2195 (CN), 1700, 1671, 1635 (C=O), 1605, 1589 (Ar-C=N, Ar-C=C); ^{1}H NMR (400 MHz, DMSO-d_6): δ_H =11.87 (bs; 1H, NH-6'), 10.55 (bs; 1H, NH-1), 8.14 (d, *J* = 2.2; 1H, H-10'), 7.79 (dd, *J* = 8.8, 2.2; 1H, H-8'), 7.48 (bs; 2H, NH_2), 7.31 (d, *J* = 8.8; 1H; H-7'), 7.18 (ddd, *J* = 7.6, 7.6, 0.9; 1H, H-6), 7.05 (d, *J* = 7.2; 1H, H-4), 6.89 (dd, *J* = 7.5, 7.5; 1H, H-5), 6.83 ppm (d, *J* = 7.7; 1H, H-7) ppm; ^{13}C NMR (100 MHz, DMSO-d_6): δ_C = 177.59 (C-2), 159.17, 158.77 (C-5',2'), 151.36 (C-10'a), 142.33 (C-7a), 136.86 (C-6'a), 134.36 (C-8'), 134.07 (C-3a), 128.38 (C-6), 124.22 (C-10'), 123.56 (C-4), 121.71 (C-5), 117.62 (C-7'), 117.31 (CN), 114.01 (C-9'), 113.24 (C-10'b), 109.24 (C-7), 107.98 (C-3a), 57.09 (C-3'), 47.74 ppm (C-3,4') ppm; ^{15}N NMR (DMSO-d_6): δ_N = 146.2 (N-6), 136.4 (N-4c), 75.1 (NH_2) ppm; MS (Fab, %): *m/z* = 435 (M+1, 28), 434 (M^+, 56). *Anal. Calcd. for* $C_{20}H_{11}BrN_4O_3$: C, 55.19; H, 2.55; Br, 18.36; N, 12.87; Found: C, 55.34; H, 2.62; Br, 18.19; N, 12.76.

2'-Amino-9'-methyl-2,5'-dioxo-5',6'-dihydro-spiro(indoline-3,4'-pyrano[3,2-*c*]quinoline)-3'-carbonitrile (3d). Colorless crystals (325 mg, 88%), M.p.: 340-2^{o}C ^{o}C; IR (KBr, cm^{-1}): $\bar{\nu}$ = 3370-3190 (NH,

NH_2), 3033 (Ar-CH), 2922 (Ali-CH), 2198 (CN), 1705, 1670, 1638 (C=O), 1610, 1587 (Ar-C=N, Ar-C=C); ^{1}H NMR (400 MHz, DMSO-d_6): δ_H = 11.83 (bs; 1H, NH-6'), 10.69 (bs; 1H, NH-1), 7.94 (s; 1H, H-10'), 7.62 (d, J = 8.4; 1H, H-8'), 7.61 (bs; 2H, NH_2), 7.43 (d, J = 8.4; 1H, H-7'), 7.35 ("t", J = 7.6; 1H, H-6), 7.18 (d, J = 7.3; 1H, H-4), 7.06 ("t", J = 7.4; 1H, H-5), 7.00 (d, J = 7.7, 1H, H-7), 2.59 ppm (s; 3H, CH_3) ppm; ^{13}C NMR (100 MHz, DMSO-d_6): δ_C = 177.86 (C-2), 159.26, 158.96 (C-5',2'), 152.23 (C-10'a), 142.37 (C-7a), 135.90 (C-6'a), 134.39 (C-3a), 132.97 (C-8'), 131.27 (C-9'), 128.23 (C-6), 123.38 (C-4), 121.64 (C-10'), 121.29 (C-5), 117.47 (CN), 115.30 (C-7'), 111.45 (C-10'b), 109.19 (C-7), 106.94 (C-4'a), 57.19 (C-3'), 47.77 (C-3,4'), 20.65 ppm (CH_3) ppm. ^{15}N NMR (DMSO-d_6): δ_N =145.5 (N-6'), 136.2 (N-1), 74.8 (NH_2) ppm; MS (Fab, %): m/z = 370 (M^+, 100). *Anal. Calcd. for* $C_{21}H_{14}N_4O_3$: C, 68.10; H, 3.81; N, 15.13; Found: C, 68.25; H, 3.99; N, 15.09.

2'-Amino-6'-methyl-2,5'-dioxo-5',6'-dihydro-spiro(indoline-3,4'-pyrano[3,2-*c*]quinoline)-3'-carbonitrile (3e). Colorless crystals (322 mg, 87%), M.p.: 310-312 ^{o}C; IR (KBr, cm^{-1}): $\bar{\nu}$ = 3359-3166 (NH, NH_2), 2193 (CN), 1711, 1672, 1645 (C=O), 1626, 1598 (Ar-C=N, Ar-C=C); NMR (DMSO-d_6) (Table 2); MS (Fab, %): m/z = 370 (M^+, 60).

Anal. Calcd. for $C_{21}H_{14}N_4O_3$: C, 68.10; H, 3.81; N, 15.13; Found: C, 68.23; H, 3.78; N, 15.21.

2'-Amino-6'-ethyl-2,5'-dioxo-5',6'-dihydro-spiro(indoline-3,4'-pyrano[3,2-*c*]quinoline)-3'-carbonitrile (3f). Colorless crystals (330 mg, 86%), M.p.: 330-332 ^oC; IR (KBr, cm^{-1}): $\overline{\nu}$ =3360-3192 (NH_2, NH), 3106 (Ar-CH), 2967 (Ali-CH), 2205 (CN), 1725, 1672, 1642 (C=O), 1600, 1578 (Ar-C=N, Ar-C=C); ^1H NMR (400 MHz, DMSO-d_6): δ_H = 10.53 (bs; 1H, NH-1), 8.09 (dd, J = 8.0, 1.2; 1H, H-10'), 7.76 ("dt", J_t = 7.2, J_d = 1.3; 1H, H-8'), 7.64 (d, J = 8.6; 1H, H-7'), 7.48 (bs; 2H, NH_2), 7.43 ("t", J = 7.6; 1H, H-9'), 7.18 ("dt", J_t = 7.6, J_d = 0.9, 1H, H-6), 7.03 (d, J = 7.2; 1H, H-4), 6.88 ("t", J = 7.5; 1H, H-5), 6.85 (d, J = 7.8; 1H; H-7), 4.14 (**A**BX_3, J_{AB} = 13.3, J_{AX} = 7.0; 1H, C**H₂**CH_3), 4.11 (A**B**X_3, J_{AB} = 13.3, J_{BX} = 7.0; 1H; C**H₂**CH_3), 1.08 ppm (AB**X₃**, J_{AX} = J_{BX} = 7.0, 3H, CH_2C**H₃**) ppm; ^{13}C NMR (100 MHz, DMSO-d_6): δ_C = 177.84 (C-2), 158.88, 158.33 (C-5',2'), 151.49 (C-10'a), 142.46 (C-7a), 137.58 (C-6'a), 134.27 (C-3a), 132.31 (C-8'), 128.29 (C-6), 123.37 (C-4), 122.66 (C-10'), 122.27 (C-9'), 121.70 (C-5), 117.44 (CN), 114.73 (C-7'), 112.49 (C-10'b), 109.21 (C-7), 106.42 (C-4'a), 57.22 (C-3'), 48.13 (C-3,4'), 36.83 (N-***C***H_2CH_3), 12.62 ppm (N-CH_2***C***H_3); ^{15}N NMR (DMSO-d_6): δ_N = 154.3 (N-6'), 136.5 (N-1), 75.1 ppm (NH_2); MS (Fab,

%): *m/z* = 384 (M^+, 100). *Anal. Calcd. for* $C_{22}H_{16}N_4O_3$: C, 68.74; H, 4.20; N, 14.58; Found: C, 68.88; H, 4.33; N, 14.41.

5. Acknowledgement

The NMR spectrometer at Florida Institute of Technology was purchased with the assistance of the U.S National Science Foundation (CHE 03 42251).

6. References

1. Obniska J, Kamiński K (2006) Acta Polon Pharm 63*:*101–108.

2. Kamiński K, Obniska J, Dybała M (2008) Eur J Med Chem 43: 53-61.

3. Obniska J, Kamiński K, Tatarcyńska E (2006) Pharmacol Rep 58: 207-214.

4. Park HB, Jo NH, Hong JH, Chei JH, Cho J-H, Yoo, KH, Oh C-H (2007) Arch Pharm (Weinheim) 340:530-537.

5. Fujio M, Hashimoto K, Katayama J, Numata A (to Welfide Corp) (2001) Preparation of spiro[azabicycloalkane-oxazolidinone] derivatives and analogs as α-7 nicotinic receptor agonists. PCT Int Appl WO 2001 066546; Chem Abstr 135:318499.

6. Pawar MJ, Burungale AB, Karale BK (2009) Arkivoc (xiii) 97-107.

7. Nakao K, Ikeda K, Kurokawa T, Togashi Y, Umeuchi H, Honda T, Okano K, Mochizuki H (2008) Nihon Shinkei Seishin Yakurigaku Zasshi (Jpn J Psychopharmacol) 28:75-83.

8. Chin Y-W, Salim AA, Su B-N, Mi Q, Chai, H-B, Riswan S, Kardono LBS, Ruskandi A, Farnsworth NR, Swanson SM, Kinghorn AS (2008) J Nat Prod 71:390-395.

9. Schick H, Frank R, Reich M, Jostock R, Bahrenberg G, Theil F, Henkel B (to Gruenenthal GmbH) (2006) Preparation of 1-oxa-2,8-diazaspiro[4.5]dec-2-enes as vanilloid receptor 1 inhibitors. PCT Int Appl WO 2006 122769 (Chem Abtrs 145:505458).

10. Hu H, Guo H, Li E, Liu X, Zhou Y, Che Y (2006) J Nat Prod 69:1672-1675.

11. Sarma BK, Manna D, Minoura M, Mugesh G (2010) J Am Chem Soc 132:5364-5374.

12. Plaska E, Aytemir M, Uzbay ÌT, Erol D (2001) Eur J Med Chem 36:539-543.

13. Pradhan R, Patra M, Behera AK, Mishra BK, Behera RK (2006). Tetrahedron 62:779-828.

14. Naeem A, Badshah SL, Muska M, Ahmad N, Khan K (2016). Molecules 21:268-286.

15. Huang D, Okada K, Komori C, Itoi E, Suzuki K (2004). Cancer Sci 95:845-849

16. Asif M, Husain A, Ahmad A, Khan AS, Rashid M, Arora K, Bahl D, Iram F (2015). Euro J Exp Bio 5:96-109

17. a) Esmaeilzadeh A, Ebteker M, Biglari A, Hassan MZ (2012). Afr J

Microbiol Res 23:4891-4896, b) Heeb S, Fletcher PM, Chhabra RS, Diggle PS, Williams P (2011). FEMS Microbiol Rev 35:247-274.

18. a) Sugar MA, Liug X-P (2000). Antimicrob. Agent 2004-2006, b) Kumar A, Fernandes J, Kumar P (2014). Int J Pharm Sci Drug Res 6:124-127, c) Hawkey MP (2003). J Antimicrob Chem 51:29-35.

19. Evans-Roberts MK, Mitchenall AL, Wall KM, Leroux J, Mylne SJ, Maxwell A (2016). J Biolog Chem, 291:3136 -3144.

20. A be M, Mai T, Ishii N, Usui M, Okuda T, Oki T (2005). Biosci Biotechnol Biochem 69:1202-1205.

21. a) Tisseh NZ, Ahmadi F, Dabiri M, Khavasi RH, Bazgir A (2012). Tetrahedron Lett 53:3603-3606, b) Lakshmi VN, Thirumurugan P, Peruma TP (2010). Tetrahedron Lett. 51:1064-1068, c) Guo Y, Li Y, Chang H, Kuo T, Han J (2016). RSC Adv 6:74683-74690, d) Auria-Luna F, Marqués-López E, Mohammadi, S, Heiran, R, Herrera PR (2015). Molecules 20:15807-15826, e) Shi R, Yan C (2016). Chin Chem Lett 27:575-578, f) Gholizadeh S, Radmoghadam K (2013). Orient J Chem 29:1637-1641, g) Wang C, Jiang Y, Yan C (2015). Chin Chem Lett 26:889-893.

22. Wenhua Chu W, Rothfuss J, d'Avignon A, Zeng C, Zhou D, Hotchkiss SR, Mach HR (2007). J Med Chem 50:3751-3755.

23.a) Wang S, Zhao Y, Bernard D, Aguilar A, Kumar S (2012). Top Med Chem 8:57-80, b) Zhang B, Golding TB, Hardcastle RI (2015). Fut Med Chem 7:631-645.

24. Sriram D, Yogeeswari P, Myneedu NS, Saraswat V (2006). Bioorg Med Chem Lett 16:2127-2129.

25. Mathusalini S, Arasakumar T, Lakshmi K, HerLin C, Mohan SP, Ramnath GM, Thirugnana-sampandan R (2016). New J Chem 1-17.

26. Yu B, Yu D, Liu H (2014). Eur J Med Chem 1-33.

27. Antonchick PA, Reimers GC, Catarinella M, Schurmann M, Preut H, Ziegler S, Rauh D, Waldmann H (2010). Nat Chem 2:735-740.

28. Aly AA (2003) Tetrahedron 59:1739-1747.

29. Hassan AA, Aly AA, El-Sheref EM (2008). J Chem. Res 9-15.

30. Mohamed AM, Alsharari MA, Aly AA (2010) J Chem Res 200-203.

31. Schulze K, Richter C, Klatt K, Ludwig R (1988) Z Chem 28:288-289.

32. Buckle DR, Cantello BCC, Smith H, Spicer BA (1975) J Med Chem 18:726-732.

33. Kaplan F, Roberts JD (1961) J Am Chem Soc 83:4666-4668.

34. Gewald, K.; Schnidler, R. *J. Prakt. Chem.,* **1990**, *332*, 223-228.

35. Sheldrick GM (2008) Acta Crystallogr A64:112-122.

36. Sheldrick GM (2015) Acta Crystallogr C71:3-8.

Figure Captions

Scheme 1. Synthesis of spiro-compounds **3a-f** *via* reaction between quinoline-2,4-diones **1a-f** and **2**.

Figure 2. Molecular structure of **3a**.DMF. 2'-Amino-2,5'-dioxo-5',6'-dihydrospiro(indoline-3,4'-pyrano-[3,2-c]quinoline)-3'-carbonitrile.DMF (1:1). Displacement parameters are drawn at 50% probability level.

Figure 3. Molecular structure of **3b**. 2 DMF. (2'-amino-9'-chloro-2,5'-dioxo-5',6'-dihydrospiro[indoline-3,4'-pyrano[3,2-*c*]quino-line)-3'-carbonitrile.2DMF. Displacement parameters are drawn at 50% probability level.

Figure 4. Molecular structure of **3c.** 2DMF. 2'-amino-9'-bromo-2,5'-dioxo-5',6'-dihydrospiro(indoline-3,4'-pyrano[3,2-c]quino-line)-3'-

carbonitrile.2DMF (1:2). Displacement parameters are drawn at 50% probability level.

Figure 5. Molecular structure of **3f.** DMF2'-Amino-6'-ethyl-2,5'-dioxo-5',6'-dihydrospiro(indoline-3,4'-pyrano[3,2-*c*]quinoline)-3'-carbonitrile.DMF (1:1). Displacement parameters are drawn at 50% probability level.

Scheme 2. Suggested mechanism for formation of products **3a-f**

Table Captions

Table 1. NMR assignment of compound **3e** ^{1}H NMR

Table 2. NMR assignment of compound **3a** ^{1}H NMR